

Synthetic Biology: A Primer

Using Genes to Make Circuits

Prabaha Gangopadhyay, IISc iGEM Team, 2016

22nd July, 2016



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- 1 Introduction
- 2 Genetic Components
 - Bacteria
 - Individual Switches
 - Circuits
- 3 iGEM
 - What is iGEM?
 - Our Project
 - Some Interesting Projects

Industrial Applications



Figure : Effluent Treatment Plant¹

¹<http://3.imimg.com/data3/HT/AQ/MY-2991599/sewage-treatment-plant-500x500.jpg>

Medical Advancements



Figure : Humulin: Synthetic Insulin²

²<http://www.humulin.com/assets/img/img-pen-vial-group-home.png>

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Bacterial Chromosome and Plasmids

- The chromosome is conserved across an entire bacterial species, but individuals in a species have different sets of plasmids.

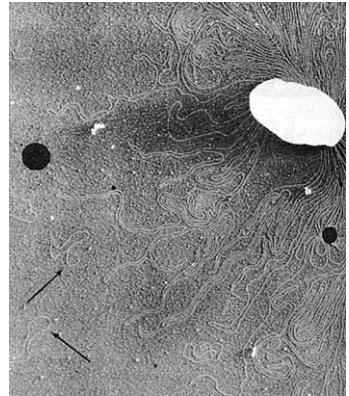


Figure : Chromosome and Plasmid³

³http://www.apsnet.org/edcenter/K-12/TeachersGuide/DNA_Easy/Article%20Images/bacterialdnax.jpg

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- The chromosome generally is several million base-pair long, while plasmids generally range in kilo-base size.

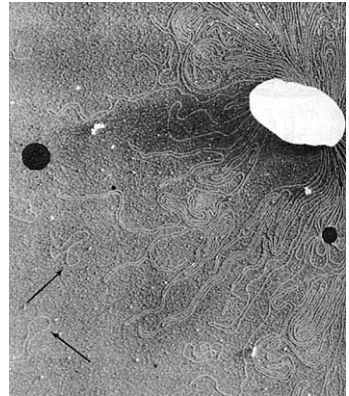


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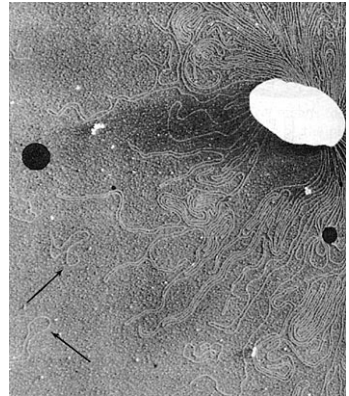


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- The chromosome generally is several million base-pair long, while plasmids generally range in kilo-base size.
- While the bacterial genome has a few thousand genes, the gene number in plasmid are in order of tens.
- Generally, plasmids do not contain genes essential for the survival of a bacteria in 'normal' conditions.

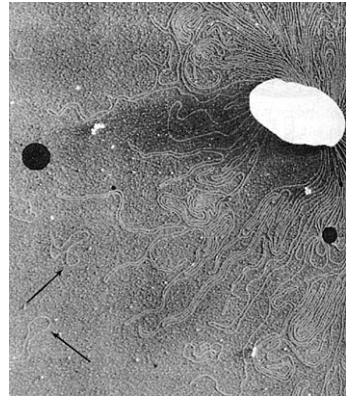


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Evolutionary Importance of Plasmids

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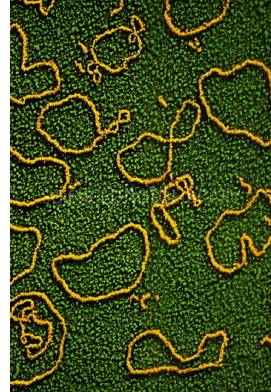


Figure : Plasmid: TEM, false-color⁴

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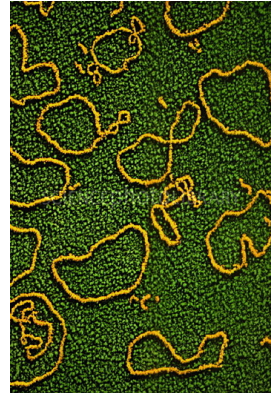


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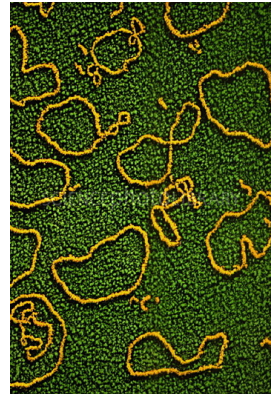


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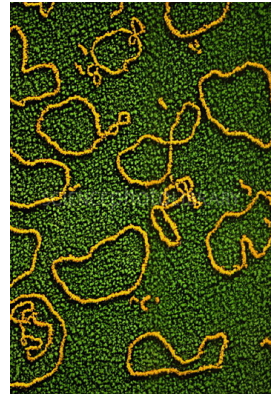


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Evolutionary Importance of Plasmids

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- Small size allows easy uptake by bacteria.
- Plays a crucial role in horizontal gene transfer.
- Helps in the process of evolution - acquiring traits!

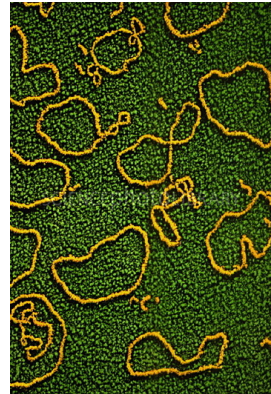


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Components of a Genetic Circuit

- **Promoter:** A nucleotide sequence that enables a gene to be transcribed. The promoter is recognized by RNA polymerase, which then initiates transcription.

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- **Inducer/Inhibitor:** A molecule that interacts with the regulator to either enhance or inhibit gene expression.
- **Operator:** A segment of DNA the regulator binds to.
- **Reporter:** A detectable polypeptide downstream of a regulon, that helps in checking the regulon activity.

Are Genes Switches?

- Genes generally are associated with a circuit, that controls it's expression.

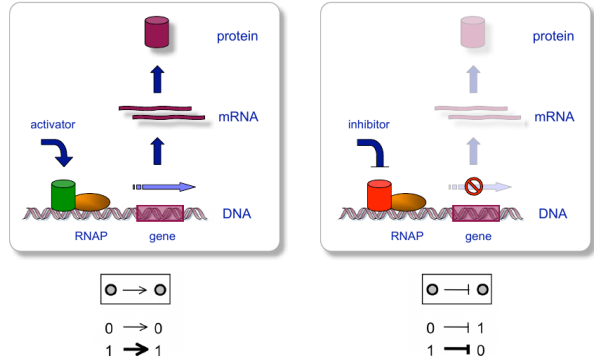


Figure : Genetic Switching⁵

⁵http://www.iiap.res.in/meet/kolleg/kollege_ppt/Thattai.pdf

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- Often, the regulation is done by two classes of molecules – inducers and inhibitors.

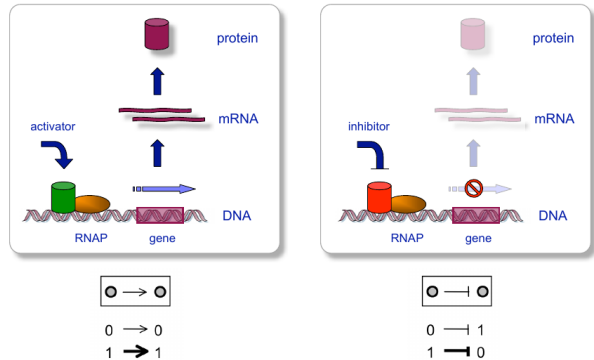


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Are Genes Switches?

- Genes generally are associated with a circuit, that controls it's expression.
- Often, the regulation is done by two classes of molecules – inducers and inhibitors.
- So, effectively, most genes can be turned on or off by the presence or absence of a molecule in the cell.

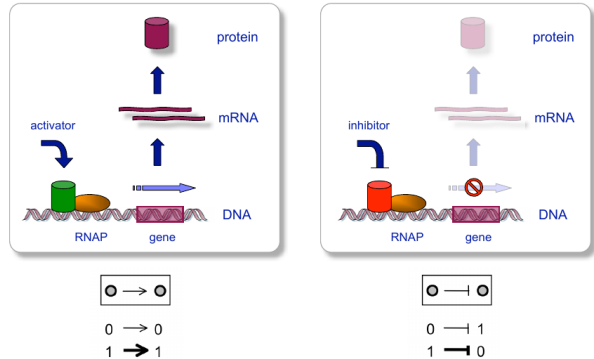


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Positive and Negative Feedback

- **Positive:** When the output of a process **activates** the process, directly or indirectly.
- **Negative:** When the output of a process **inhibits** the process, directly or indirectly.

Quorum Sensing: Positive Feedback

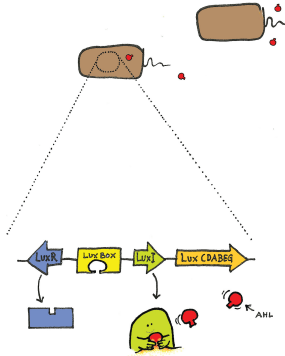


Figure : Low Bacterial Population⁶

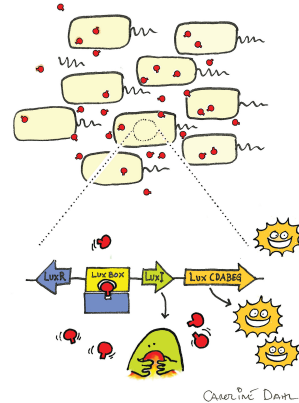


Figure : High Bacterial Population⁶

⁶https://upload.wikimedia.org/wikipedia/commons/b/b2/Bacterial_Quorum_Sensing_by_CarolineDahl.jpg

Planarial Wound Healing: Negative Feedback

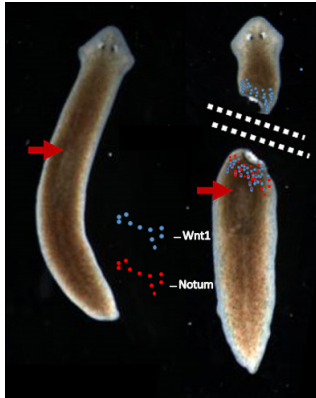


Figure : Wound Healing⁷(Edited)

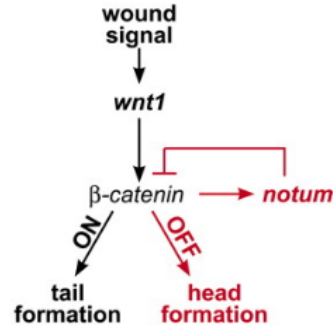


Figure : The Pathway⁸

⁷<http://voices.nationalgeographic.com/files/2013/07/Figure-4-piece.jpg>

⁸Petersen, Christian P., and Peter W. Reddien. Science 332.6031 (2011): 852-855.

Some Simple Genetic Circuits

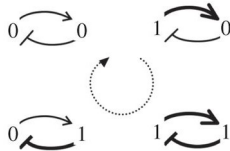
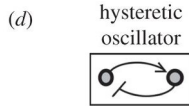


Figure : Hysteric Oscillator⁹



Figure : Flip Flops⁹

⁹Thattai, Mukund. Philosophical Transactions of the Royal Society of London A: Mathematical, Physical and Engineering Sciences 371.1984 (2013): 20110548.

Repressator: A Slightly Complex Circuit

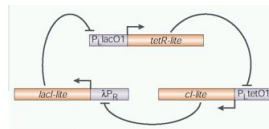
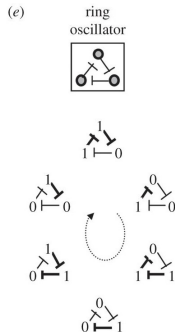


Figure : Repressator¹¹

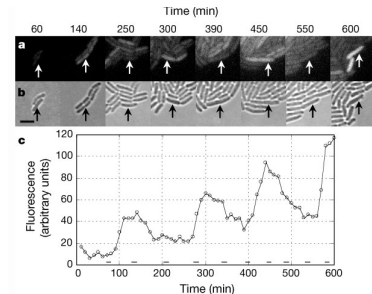


Figure : Repressator Output¹¹

Figure : Ring Oscillator¹⁰

¹⁰Thattai, Mukund. Philosophical Transactions of the Royal Society of London A: Mathematical, Physical and Engineering Sciences 371.1984 (2013): 20110548.

¹¹Elowitz, Michael B., and Stanislas Leibler. Nature 403.6767 (2000): 335-338.

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Is Bacteria a Machine?

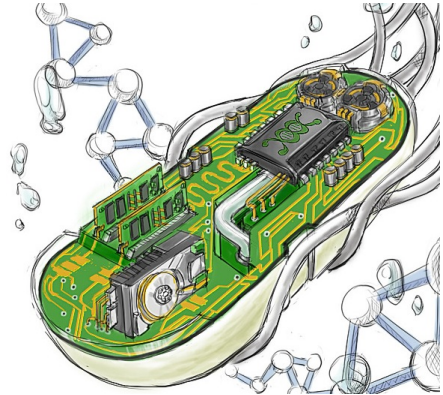


Figure : *E. coli*: The Prokaryotic Lab Rat¹²

¹²<http://www.rle.mit.edu/sbg/wp-content/uploads/2013/03/O3-1-colors.jpeg>

Is Bacteria a Machine?

- A system analogous with an autonomous motherboard, with some replaceable ICs, to modify output, exploiting the existing circuitry.

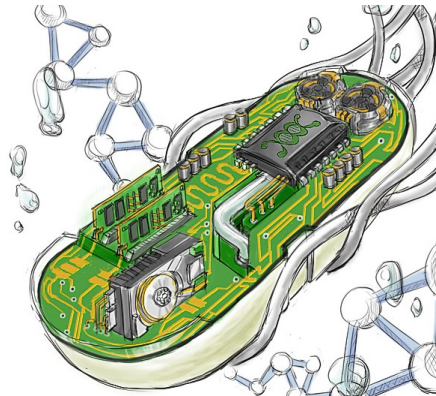


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- The plasmids are the ICs.

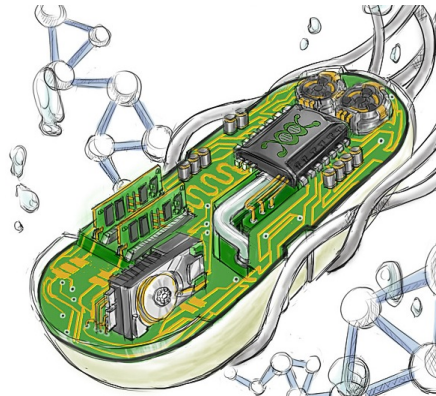
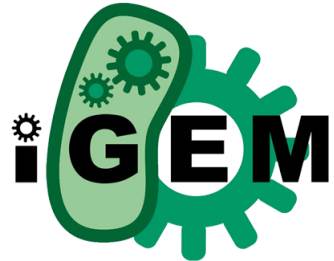


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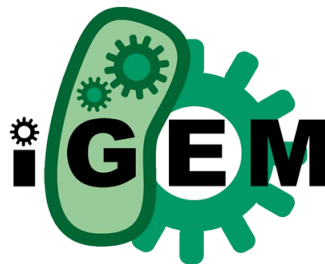
iGEM

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- There are a few more technical details, but they are not particularly necessary.



Cellfuge: IISc Bangalore, 2016

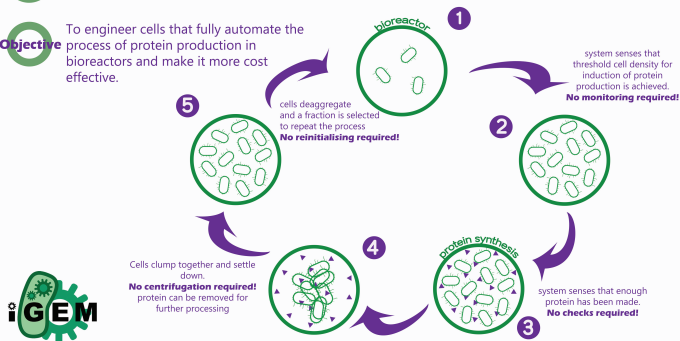
Cellfuge



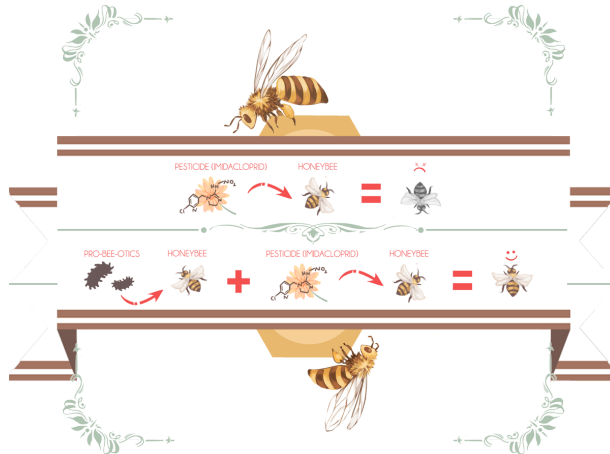
iGEM is a synthetic biology competition that aims at constructing biological solutions to everyday problems using standard parts.



To engineer cells that fully automate the process of protein production in bioreactors and make it more cost effective.

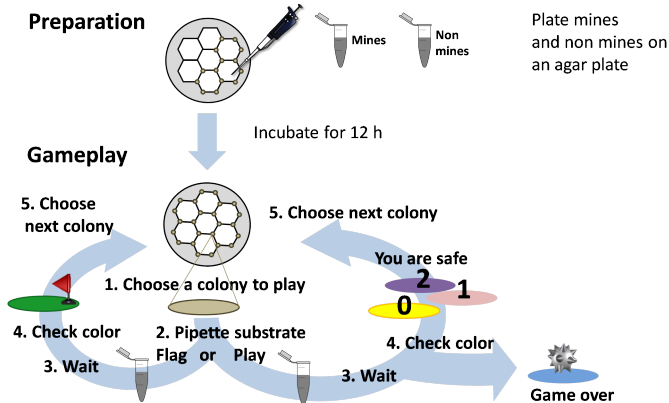


Probeeotics: British Columbia, 2015



Colisweeper: ETH Zurich, 2012

How to play Colisweeper?



Acknowledgements

- Prof. Deepak Saini, MRDG, IISc
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- Dr. Sreenath Thiruneelakantan, Instructor, UG Program, IISc
- Chaitra Prabhakara, PhD Student, NCBS
- My teammates: Abhijeet, Aiswarya, Aneesh, Arunavo, Ayan, Jessica, Sandhra, Shreyas